STUDY OF HEMATOLOGICAL PROFILE IN

RHEUMATOID ARTHRITIS PATIENTS

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CERTIFICATE

This is to certify that this dissertation titled "**STUDY OF HEMATOLOGICAL PROFILE IN RHEUMATOID ARTHRITIS PATIENTS"** submitted by **DR. R.ARUL** to the faculty of General Medicine, **The Tamil Nadu Dr. M.G.R. Medical University, Chennai** in partial fulfillment of the requirement for the award of MD degree branch I General Medicine, is a bonafide research work carried out by him under our direct supervision and guidance.

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DECLARATION

I**, Dr.R.Arul,** solemnly declare that the dissertation titled "**Study of hematological profile in rheumatoid arthritis patients"** has been prepared by me.

This is submitted to **The Tamil Nadu Dr. M.G.R. Medical University, Chennai,** in partial fulfillment of the rules and regulations for the award of MD degree (branch I) General Medicine.

Place: Madurai

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CONTENTS

INTRODUCTION

Rheumatoid Arthritis (RA) in this commonest form of chronic inflammatory joint disease. It is a symmetrical, non supportive polyarticular disease unique to modern man.[1]

Rheumatoid arthritis affects the synovial joints, but it is not confined to them and the many visceral manifestations have led to the classification of RA as a systemic disorder of the immunological mechanism.

Of the systemic lesions, anemia and a focal subcutaneous granuloma are the most characteristic. Some cases of RA, particularly those that are seropositive, are fulminating and rapidly progress to severe deformity.[1]

REVIEW OF LITERATURE

HISTORICAL REVIEW

Historical review revealed that Hippocrates and other Greek and Roman writers gave possible descriptions of the disease (Short, 1974). There are suggestive descriptions in the Sanskrit writings of Charaka Samhita of 100 AD (Sturrack *et al.* 1977) and by the 17th century English Physician, Thomas Sydenham (short 1974).

Sydenham described the diseases state which he called "Rheumatoid Polyarthritis". But it was clearly described by landre-Beauvais (1800). Garrod (1859) used the term "Rheumatoid Arthritis" and a clinical description of the disease entity began to emerge. The modern concept of the disease evolved by the pain-staking work of Nicholes and Richardson (1906), Jones, Milard Smith (1930), Lewis Faning, Short, Bauer and Reynold (1957). Short's text book (1957) set the stage for the modern era of investigation and inquiry in the field of clinical rheumatology. American Rheumatism Association carefully drafted the description and the criteria for the diagnosis of Rheumatoid Arthritis.

DEFINITION

Rheumatoid Arthritis is a chronic, symmetrical, inflammatory, deforming polyarthritis affecting small and large peripheral joints with associated systemic disturbance such as vasculitis and nodules. Characteristically the course of the disease is prolonged with exacerbations and remissions. [2]

GENERAL ASPECTS OF RHEUMATOID ARTHRITIS

EPIDEMIOLOGY

Rheumatoid Arthritis occurs throughout the world and in all ethnic groups. Its prevalence is approximately 1 percent of the population (range 0.3 to 2.1 percent); women are affected approximately three times more often than men. The prevalence increases with age and sex differences diminish in the old age group. The onset is more frequent during the fourth and fifth decade of life, with 80% of all patients developing the disease between the ages of 35 and 50. [(2)]

AETIOLOGY

Rheumatoid Arthritis is a disease determined by genetic and environmental factors, namely an infectious agent which is systemically distributed in the patient, but has a particular predilection for synovial joints.

GENETIC FACTORS

Family and mono and dizygotic twin studies have shown that there is a genetic factor in Rheumatoid Arthritis. Severe Rheumatoid Arthritis is found at approximately four times the expected rate in first degree relative of individual with seropositive disease. [(2)]

Moreover 30% of monozygous twins are concordant for Rheumatoid Arthritis whereas only 5% of dizygous twins are concordant.

In Rheumatoid Arthritis the important observation was the finding that HLA-D4 and HLA-DR4 are significantly elevated, being found in 60-80% of patients compared to 20% of control subjects. (Panayi *et al,* 1978; Stastny, 1978). This is true for all populations except Israel & India where the association is with HLA-DR1 (Schiff *et al*, 1982).

Environmental Factor

Increasing urbanization and industrialization in Europe from 18th century onwards led to the spread of rheumatic diseases. Such a demographic change with a change in the prevalence of Rheumatoid Arthritis seems to have been observed in South Africa.

The urban factor involves alteration in diet, living conditions and exposure to new environmental agents both microbial and chemical. Indirect evidence for such environmental factor comes from the work of Wasmuth *et al* (1972) who showed that the probability of the non-consanguineous spouse of a proband with Rheumatoid Arthritis developing the disease was related to the length of time the two individuals had lived together.

INFECTION-PATHOGENESIS

Rheumatoid Arthritis is characterized by persistent cellular activation, autoimmunity and the presence of immune complexes at sites of articular and extra-articular lesions.

Evidence suggesting RA as autoimmune disease[1]

1) Hypergammaglobulinemia
2) Presence of variety of auto antibodies especially Rheumatoid Factor.
3) Presence of circulating and deposited immune complexes.
4) Cryoglobulins.
5) Plasma cells in bone marrow
6) Central lymphoid organs and synovial membrane making rheumatoid factor.
7) Infiltration of the synovial membrane with mononuclear chronic inflammatory cells, especially lymphocytes.

The central question concerns the mechanism whereby the autoimmunity is switched on. There are those who postulate that initiation of the disease is a genetically determined failure to IgG while others maintain that external agents or factors activate autoimmunity to IgG by the breaking of tolerance through a variety of mechanisms such as excessive helper activity, loss of suppressor activity and activation of

macrophages (Carson, 1982). Recent experimental work has demonstrated that the production of antiglobulins and antinuclear factors is a very prominent component of a normal immune response although the physiological function of such auto antibodies is not known. (dresser, 1978; Steele & Cunningham, 1978).[1]

In Rheumatoid arthritis there is evidence of abnormal immune reactivity in the bone marrow, lymph nodes and spleen as well as in the synovial membrane itself.

As a consequence of the persistence of the Rheumatoid Arthritis Agent and the consequent chronic stimulation of the immune response, a variety of auto antibodies are formed and in particular rheumatoid factor. These factors and their ability to form immune complexes are thought to be responsible for some of the inflammatory lesions especially the extraarticular features, seen during the course of the disease. Some of the immune complexes formed locally in the joint while others may be formed in the circulation with subsequent deposition in the tissues.

Disease in the joints

The earliest lesion in Rheumatoid Arthritis involves the blood vessels running through the synovium, followed by the perivascular accumulation of lymphocytes and lastly by changes in the synovial lining cell layer.

This leads to swelling and congestion of the synovial membrane and the underlying connective tissues, which become infiltrated with CD4 T cells, macrophages and plasma cells. Some 70% of the latter are producing IgG and IgM rheumatoid factors. Immune complexes are present in abundance both extra cellularly and intracellularly either as phagocytosed material or as self associated IgG rheumatoid factors within the plasma cells (Munthe, 1978).

Effusion of synovial fluid into joint space takes place during active phases of the disease. Hypertrophy of the synovial membrane occurs with the formation of lymphoid follicles resembling an immunologically active lymph node. Inflammatory granulation tissue (Pannus) is formed, spreading over and under the articular cartilage which is progressively eroded and destroyed.

Later, fibrous adhesions may form between the layers of pannus across the joint space and fibrous or bony ankylosis may occur. Muscles

adjacent to inflamed joints atrophy and there may be focal infiltration with lymphocytes.

Extra Articular Diseases

The most frequent tissue lesion is the subcutaneous granulomata. It is histologically more characteristic (Moore & Wilikens, 1977). These are found in 20% cases, usually seropositive.

Subcutaneous nodules have a characteristic histological appearance. There is a central area of fibrinoid material consisting of swollen and fragmented collagen fibers, fibrinous exudates and cellular debris, surrounded by a palisade or radically arranged proliferating mononuclear cells. The nodules have a loose capsule of fibrous tissue.[3]

Similar granulamatous lesion may also occur in the pleura, lung, pericardium and sclera. Lymph nodes are often hyperplastic, showing many lymphoid, follicles with large germinal centers and numerous plasma cells in the sinuses and medullary cords. Immunofluorescence shows that plasma cells in the synovium and lymph nodes synthesize rheumatoid factors.

The 1987 Revised Criteria for the Classification of Rheumatoid Arthritis[4]

I. Guideline for Classification

Four of seven criteria are required to classify a patient as having Rheumatoid Arthritis.

II. Criteria

1. Morning stiffness – stiffness in and around the joints lasting one hour before maximal improvement.

2. **Arthritis of three or more joint areas.**

   At least three joint areas, observed by a physician simultaneously, have soft tissue swelling or joint effusions, not just bony growth. The 14 possible joint areas involved are right or left proximal interphalangeal, metacarpophalangeal, wrist, elbow, and knee, ankle, and metatarsophalangeal joints.

3. **Arthritis of hand joints**

   Arthritis of wrist, metacarphophalangeal joint, or proximal interphalangeal joint.

4. **Symmetric arthritis**

Simultaneous involvement of the same joint areas on both sides of the body.

5. **Rheumatoid Nodules**

Sub cutaneous nodules over bony prominences, extensor surfaces, or juxtaarticular regions observed by a physician.

6. **Serum rheumatoid factor**

Demonstration of abnormal amounts of serum rheumatoid factor by any method for which the result has been positive in less than 5% of normal control subjects.

7. **Radiographic changes**

Typical changes of Rheumatoid Arthritis on poster anterior hand and wrist radiograph which must include erosions or unequivocal bony decalcification localized in or most marked adjacent to the involved joints.

*Criteria 1-4 must be present for atleast 6 weeks. Criteria 2-5 must be observed by a physician.

Articular Manifestations of Rheumatoid Arthritis

The onset of the disease is usually insidious especially in the 4th and 5th decades. Occasionally the onset may be acute as in Still's disease with high grade fever, evanescent rash, leucocytosis, lymphadenopathy, spleenomegaly, pleuritis and pericarditis (Bywaters, 1971; Esdaile *et al* 1980). More commonly the arthritis is polyarticular and symmetrical from the outset (Hernandez *et al* 1980). The test for rheumatoid factor is usually positive at this state (Jacoby *et al* 1973). The traditional view is that small peripheral joints are affected first, but in one large study large joints were affected first in 52% of patients (Short *et al* 1957).

Regarding prognosis, as per one study, upto one quarter of the patients had inactive disease and functional ability and another quarter had only mild to moderate activity with only slight functional impairment. The remaining patients, however, had persistent disease activity and were severely crippled (Short and Bauer 1948; Ragan 1949; Short *et al* 1957; Duthie *etal* 1964).

Patients with persistently high titers of rheumatoid factor have in general a poor prognosis, whereas those with low titers or intermittently weakly positive rheumatoid factor tests run a favorable course (Duthie *et al*, 1964; Feigenbaum *et al,* 1979).

The clinical features of the arthritis conform to the cardinal features of inflammation with the exception-redness is never present. A red rheumatoid joint always must be suspected as being infected usually with staphylococcus aureus and demands immediate arthrocentesis. (Kellgren et al 1958; Karten 1971; Huskisson and Hart 1972).

Joint stiffness is especially troublesome in the morning, attributed to increased fluid content of the joint tissues (Scott 1960). Stiffness after sitting for some time is also very characteristic of rheumatoid arthritis, although also found in other forms of inflammatory joint disease (Wright and Plunkett, 1966).

The joints most frequently involved are the proximal interphalangeal joints of the fingers, the interphalangeal joints of the thumbs, the metacarpophalangeal joints, the wrist, elbow and shoulder joints, the knee joints and metatarsophalangeal joints in the feet. The pattern of joint involvement is so often symmetrical (Jacoby *et al* 1973; Fleming et al 1976 a, b) typically the distal interphalangeal joints are spared.[4]

Involvement of the temporomadibular joints and the cervical spine are commonly present as early features in the disease presentation

(Fleming *et al* 1976). Severe disease is generally associated with early wrist and metatarsophalangeal joint involvement.

Clinical Features of the Joints of Hand and Wrist in Rheumatoid Arthritis

Dorsal Surface

1. Synovial Hypertrophy of the wrist joint.
2. Extensor tendon and extensor pollicis longus involvement.
3. Prominence of the ulnar styloid.
4. Wasting of the dorsal interossei
5. Involvement of the metacarpophalangeal joints with ulnar deviation.
6. Involvement of the proximal interphalangeal joints with 'Buttonhole' and 'Swan-neck' deformities.
7. The typical z-shaped deformity of the thumb.

Volar Surface

1. Swelling along flexor sheaths and 'trigger finger'
2. Heaping up of the flexor retinaculam, with compression of median and ulnar nerves.
3. Wasting of intrinsic muscles due to median or ulnar nerve compression or peripheral neuropathy.

Systemic Manifestations of Rheumatoid Arthritis

The commonest non-articular manifestation of Rheumatoid Arthritis in the granulomatous nodule, which is characteristically found near the olecranon process in 20 to 30% of patients with 'definite' or 'classical' Rheumatoid Arthritis as defined by the American Rheumatism Association. (Ropes *et al*, 1959). It can also occur over scalp, sacrum, scapula, Achilles tendon, fingers and toes.

Vasculitis – Arterial inflammation is often clinically silent, but when severe and extensive may cause dermal infarction, peripheral neuropathy, perforation or gangrene of the bowel and even myocardial infarction**.** (Webb & Payne, 1970; Lind – say *et al*, 1973). Approximately 5 to 10% of all Rheumatoid Arthritis patients, especially females will develop leg ulcers at some time of their illness.

In respiratory system, pulmonary involvement is important. Caplan syndrome consists of nodular opacities varying in size from 0.5 to 5 cm throughout both lungs in a patient with Rheumatoid Arthritis or in a subject with minimal or no Rheumatoid Arthritis but with positive tests for rheumatoid factor (Miall *et al,* 1953; Caplan *et al*, 1962**).** Interstitial fibrosis, pleurisy, pleural effusion and pleuropulmonary nodules can also occur.

Heart involvement in Rheumatoid Arthritis include granulomatous infiltration of myocardium and conducting system, valvular insufficiency (Carpenter *et al*, 1967) pericarditis and myocardial failure.

In eye keratoconjunctivitis sicca occurs in about 10% of patients with Rheumatoid Arthritis (Bloch *et al,* 1965). Episcleritis and Scleritis also occur.

Sjogren syndrome comprises the triad-dry eyes of keratoconjunctivitis sicca, xerostomia with or without salivary gland enlargement and in one-half to two-third of patients a connective tissue disease, usually Rheumatoid Arthritis. (Bloch *et al,* 1965; Whaley *et al,* 1973 a, b).

Neurological manifestations may result from atlantoaxial or midcervical spine subluxations, entrapment neuropathy of median, ulnar, radial or anterior tibial nerves, mononeuritis multiplex and peripheral neuropathy.

Osteoporosis, muscle weakness and wasting may occur adjacent to inflamed joints.

Hematological involvement in rheumatoid arthritis

The most common extra articular abnormality seen in patients with rheumatoid arthritis is anemia. This is usually a mild anemia that is either symptomatic or accompanied by slight fatigue. (Katyrin *et al* 1987).

Many patients with rheumatoid arthritis have an anemia which responds poorly to standard hematinic therapy. (Mowat *et al* 1971). Fortunately the degree of anemia is usually low and there is correlation between the severity of disease and depth of anemia (Grennan *et al* 1975).

Anemia in rheumatoid arthritis:

1. Anemia of chronic disease
2. Iron deficiency anemia due to

   (a) poor diet

   (b) Chronic blood loss due to treatment

   Salicylate-chronic blood loss

   Phenacetin - Methhemoglobinemia

   Heinz body hemolytic anemia

   Phenyl butazone-pancytopenia

   Gold-thrombocytopenia and pancytopenia

   (c) Significant bleeding in to the joint
3. Folate, B12 deficiency due to drugs[5]

Anemia of chronic disease is present in 27% of outpatients followed for rheumatoid arthritis [5] and 50% of newly diagnosed patients admitted in wards. Anemia of chronic disease is normocytic or normochromic or slightly hypochromic. Bone marrow is usually normal except for slight under hemoglobinisation of mature precursors. [6], Iron stores are normal but sideroblasts are decreased.

	Anemia of chronic Disease	Iron deficiency anemia	Fedeficiencywith Inflammation
MCV	72-100fl	<85fl	<100fl
MCHC	<36	<32	<32
Serum iron	Decreased	decreased	Decreased
TIBC	Below mid normal Range	increased	Less than upper limit of normal
Transferrin sat.	>20%	<15%	15%
Serum ferritin	>35ng/ml	<35ng/ml	<200ng/ml
Serum soluble transferring Receptor conc.	Normal	increased	Increased
Stainable iron in bone marrow	+	—	—

The pathogenesis of anemia in rheumatoid arthritis is complex. (Mowat *et al* 1971)

Chronic administration of anti rheumatic drugs can lead to GI blood loss. (Scott *et al* 1961)

There is evidence that the incidence of macrocytic anemia is higher than expected in patients with rheumatoid arthritis largely owing to folate deficiency (Gough *et al* 1964)

The pathogenesis of anemia in rheumatoid arthritis

(1) Shortened red cell survival (Dinant *etal* 1978, Richmond *etal* 1978)

(2) Reduced serum and red cell folate concentration (Omer 1968)

(3) Impairment of compensatory increase in iron absorption in response to anemia(Boddy & will 1969)

(4) Improved erythropoietin release in response to anemia (Ward *et al* 1969)

(5) Increased ineffective erythropoiesis(Dinanat *etal* 1978)

(6) Synovial iron deposition (Muirden and senator 1967)

(7) An abnormal pattern of iron handling characterized by rapid serum iron release and defective release of stored iron from reticuloendothelial system(Owen& Lawson1966, Bennet *etal* 1974)

(8) Suppression of proliferative capacity of erythroid bonemarrow by immune mechanisms by both humoral (Dainak *etal* 1980, Goldberg et al 1980)and cell mediated mechanism (Abodu *et al* 1978)

Microcytosis <80 fl present in 2 to 8% anemia of chronic disease Most recent series states that it is present in 20 to 40%[7,8]of patients with rheumatoid arthritis.

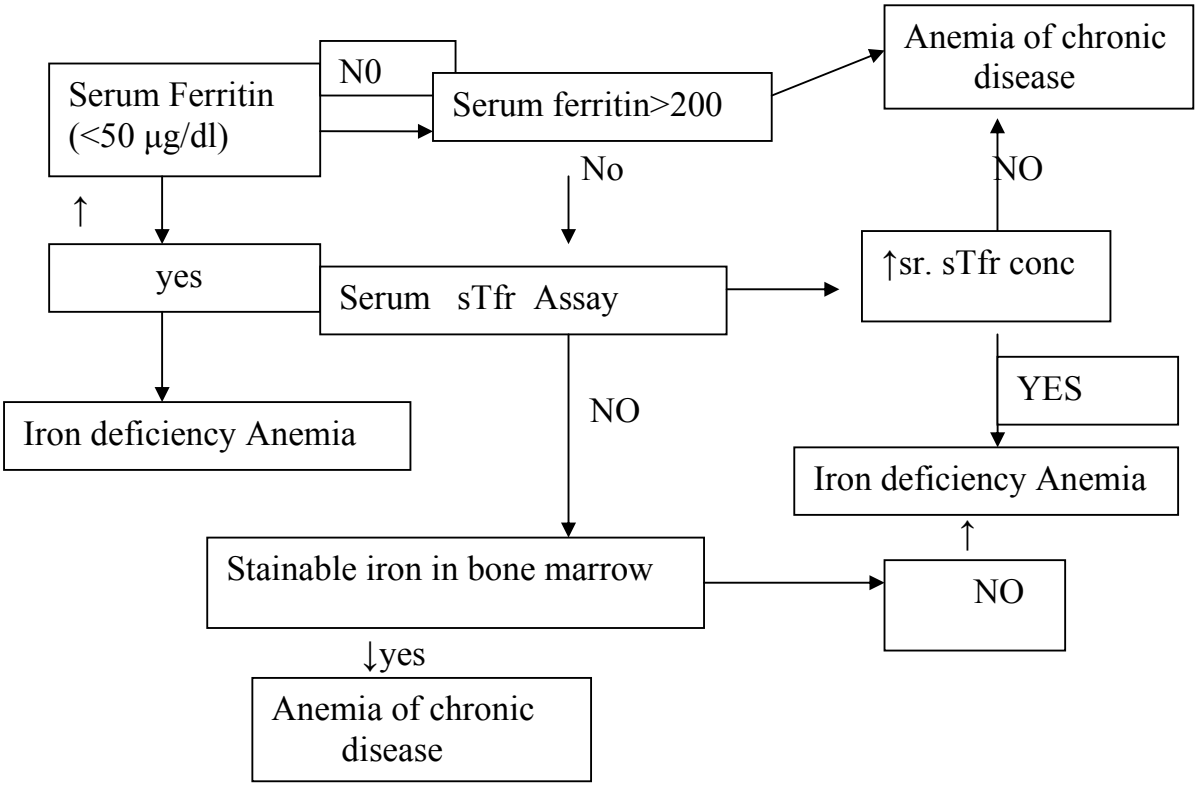
Hypochromia (MCH < 26) is more common than microcytosis. It is present in 50-100% of patients with rheumatoid arthritis .Hypochromia is observed even though hematocrit remains normal .

MCV<72fl is rare [9]

RDW is typically elevated [9]

Hypochromia typically precedes microcytosis in anemia of chronic disease but typically follows the development of microcytosis in iron deficiency.

Algorithm for evaluation of anemia of chronic disease:



Mechanism for anemia of chronic disease:

(1) Shortened red cell survival [10,11]

(2) Impaired marrow response

(a)Impaired erythropoietin production

(b)Impaired marrow progenitor response to erythropoietin

(3)Impaired mobilization of RE iron stores

Shortened red cell survival

Anemic patients with rheumatoid arthritis show inverse correlation between IL-1 level and red cell survival. [12, 13] There is a decrease in erythropoietin availability resulting in selective hemolysis of youngest red cells. Another mechanism described is increased generation of peroxy nitrite in red cells may enhance membrane rigidity and shortened red cell survival. The mean survival of RBC survival is 80 to 90 days compared to 100 to 120 days in normal persons. in rheumatoid arthritis.[13,14]

Impaired erythropoietin production

There is an inverse correlation between serum erythropoietin level and hemoglobin, as hemoglobin decreases erythropoietin increases. For any given anemic individual with RA patients, erythropoietin level was lower than that found in equally anemic individuals with iron deficiency. Thus erythropoietin response to anemia was blunted in RA[15, 16, 17, 18]. Impaired Erythropoietin response is due to cytokines like IL1, TNF α, TGF β which inhibit the production of erythropoietin. [5]

Impaired marrow response to erythropoietin:

Although erythropoietin levels of patients with ACD are not high as those in equally anemic patients with iron deficiency, these values are still higher than normal individuals who are not anemic. Other factors like TNFα, IL-1, IFNγ also inhibits erythroid colony formation.

TNF α was found to inhibit colony formation by erythroid colony forming units from BM mononuclear cells in a dose dependent fashion[19)]. However colony formation by highly purified CFU-E generated from peripheral blood cells (300CFU-E) was not affected; indicating that TNFα action was indirect, which is likely to be mediated by marrow stroma. Inhibition was mediated by soluble factors released from stroma, including IFNγ [20]. Similarly inhibitory effect of IL-1 on CFU- E colony formation was also shown to be indirect and dependent on IFNγ released from T lymphocytes. IFN γ induces apoptosis in CFU-E a process that requires Fas activation .Ceramide , a product of sphingomyelin hydrolysis, a mediator of apoptotic effects of TNFα,IL1,IFN γ and is frequently implicated in Fas mediated events.It is either endogenous ceramide produced by exposure to bacterial sphingomyelinase or exogenous bacterial sphingomyelinase or exogenous cell permeable ceramide which significantly inhibits bonemarrow CFU- E derived colony formation. Exposure of marrow cells to IFN y lead to significant increase in ceramide content suggesting a role of ceramide. Nitric oxide,

another potential 2nd messenger in cytokine effects, directly inhibits erythroid colony formation in vitro .Cytokines may alter the expression of hematopoeisis growth factor receptors during erythroid development as well. Exposure to 2500 u/ ml IFNγ in vitro resulted in a decrease in erythroid receptors for EPO and stem cell factor but not insulin like growth factor 1 probably mediated at the gene translation level.

Impaired mobilization of Reticuloendothelial iron store

Impaired mobilization of reticuloendothelial iron store is also from cytokine effects.IL1 increase the translation of ferritin messenger RNA [21, 22)] and additional ferritin could act as a trap for iron that might otherwise be available for erythropoiesis. The acute phase reactant α_1AT is inhibiting erythropoiesis impairing transferring binding to transferring receptor and subsequent internalization of Tfr

Increase serum concentration of soluble transferring receptor may able to identify iron deficiency. [23,24] Concentration of free protoporphyrin in erythrocytes tend to be elevated in patient with anemia of chronic disease. FEP increases more slowly in anemia of chronic disorder than it does in iron deficient and it does not become clearly abnormal with significant anemia has developed. Hepcidin is a acute phase reactant, anti microbial protein , regulator of iron storage and transport and has been proposed as a mediator of anemia of chronic disease. Recent studies have

underlined the role of hepcidin, a peptide produced by hepatocytes, in the pathogenesis of ACD. Interleukin-6 produced at sites of inflammation or infection stimulates hepcidin production, an acute-phase protein, which causes sequestration of iron in reticuloendothelial cells and also decreases intestinal iron absorption [25]. Hence blocking hepcidin production may help in effective treatment of ACD.

Eosinophilia

Normally eosinophils are present upto 5% of total WBC. Eosinophilia occurs in 40% in rheumatoid arthritis patients .(26)

Large granular lymhocytosis syndrome

The subset of patients with rheumatoid arthritis has increased number of large granular lymphocytes in peripheral blood, bone marrow and liver. The lymphocytes contain many azurophilic granules in cytoplasm and may account for 90% mononuclear cells . In LGL, 1/3 rd have Rheumatoid arthritis . LGL syndrome in patients with rheumatoid arthritis has same HLA DR 4 association as seen in Felty syndrome. (27)

Platelet abnormalities

Thrombocytosis is a common finding in acute and severe rheumatoid arthritis. .(28) It correlates with most parameters of disease

activity such as ESR, WBC count, plasma viscosity, liver enzymes, rheumatoid factor and acute phase reactants (Farr *et al* 1983).

Thrombocytopenia is not a feature of rheumatoid disease except in Felty syndrome or as a result of gold and D penicillamine therapy. Shortened platelet survival is found in a minority of patients with rheumatoid disease but these patients usually have normal or increased platelet count (Farr *et al* 1980; kelton *et al* 1983).

In patients with platelets greater than 10,00,000, an alternative causes of the thrombocytosis should be considered.

NEUTOPENIA

In 1924 Felty described the clinical association of leucopenia, splenomegaly, and rheumatoid arthritis. Other features include lympadenopathy, weight loss, anemia, skin pigmentation, chronic leg ulceration. It occurs in about 1% of patients with rheumatoid arthritis.[29]

The pathogenesis of neutropenia is multifactorial. The following have been described.

1. Antineutrophil antibodies
2. Antigen-antibody complexes
3. Myelosuppression by a humoral inhibitor as well as a mononuclear suppressor cell **(**Goldberg & pinals 1980;logue *etal* 1981; backnall *et al* 1982)

Hyperviscosity syndrome:

Patients with high titres of rheumatoid factor have slightly increased serum viscosity. Occasionally this may be marked and give rise to hyperviscosity syndrome. (Pope *et al* 1975; Cryer *et al* 1981**)**

The clinical features consist of dyspnoea, dizziness, ataxia diplopia, visual blurring, epistaxis, skin hemorrhages, menorrhagia and rectal bleeding. Examination of fundus reveals venous engorgement , blot hemorrhages, soft exudates and in severe cases papilloedma.

Paraproteinemia

It is typified by monoclonal gammopathies and it has poor prognostic significance when it appears in rheumatoid arthritis patients[30]

Hematological malignancies:

There are conflicting reports on the risk of lymphoma and leukemia in patients with rheumatoid arthritis. The bulk of evidence suggests that the risk is increased. In review of 46,101 patients with rheumatoid arthritis, 130 cases of leukemia, lymphoma and myeloma were observed as compared to 59.6 cases expected in general population, which is statistically significant . [(31)]

There is an increased risk of hematologic malignancies in patients with Sjogrens syndrome and probably in those with rheumatoid arthritis. In Sjogrens syndrome the incidence of non Hodgkin's lymphoma is more than 40 times of what is expected in the general population.

Coagulation abnormalities:

Conn *et al* 1976[32] found that elevated level of fibrinogen and FDP in patients with rheumatoid vasculitis. C.S Lau etal in 1993 found that VWF is elevated in patients with rheumatoid vasculitis. Pariaz *et al*[33] states that some patients develop acquired "factor VIII inhibitors" and acquired hemophilia in rheumatoid arthritis patients .In their study one patient had decreased protein S and protein C .

AIMS AND OBJECTIVES

(1) To study the hematological status in patients with rheumatoid arthritis.

(2) To find out the prevalence of anemia in these patients and its correlation with seropositivity and disease activity which is measured by DAS 28 score(Disease activity score).

(3) To know the prevalence of iron deficiency anemia and anemia of chronic disease among anemic patients of rheumatoid arthritis and its correlation with disease activity i.e DAS 28 score.

(4) To analyse other hematological parameters and its correlation with DAS 28 score.

Materials and Methods

Setting	: Department of Medicine , Govt Rajaji hospital
Design	: Cross-sectional study
Period of study	: One year study
Ethical approval	: Obtained from ethical committee approval headed by Dean, Govt. Rajaji hospital
Consent	: Obtained from all patients
Statistical software	: EPI Info 2002
Study population	: Patients attending Rheumatology OP- randomly selected

Inclusion criteria:

(1)Patients who satisfied the American Rheumatologic association criteria 1987, irrespective of hematological signs present or not .

(2)Age group 20 to 60 years irrespective of any sex.

(3) duration of disease upto 5 years

Exclusion criteria:

(1)Previously diagnosed anemia and treated

(2)Previously have any other bleeding disorder not related to Rheumatoid arthritis

(3)Those who have mixed disorder like SLE and RA ;SS & RA and MCTD and overlap syndrome.

(4)previously knowm malignancies, renal failure, hemolytic anemia

(5) any other chronic blood loss like hemorrhoids

Forty four patients of rheumatoid arthritis were selected from random basis for the study from rheumatology clinic , Govt. Rajaji hospital, Madurai.

The duration of illness was upto 5 years at different stages of rheumatoid arthritis. Patients with the age group ranging from 20 to 60 years were studied.

The selected patients were evaluated with detailed history regarding duration of disease and history of drug intake and type of onset of symptoms noted.

Presence of joint swelling, tenderness and deformities and number of tender joints and number of joint swelling noted. Rheumatological functional class was assigned clinically.

Detailed clinical examination including pallor and rheumatoid nodule and lymphadenopathy.All systems are examined carefully and visual analogue pain score was carefully assessed.

Hemoglobin, Red blood cell count, White blood cell count , Hematocrit,Differential count , MCV , MCH, MCHC , Platelet count , RDW, MPV, Serum ferritin, Total iron binding capacity, Serum iron, Peripheral smear, , Bleeding time , Clotting time, ESR, Rheumatoid factor, C reactive protein , Sugar, urea , creatinine, serum protein , albumin and globulin were done in laboratory.

Automated hemogram was done to calculate blood cell counts through 3 part differential automated coulter cell counter.

Most automated counters measure hemoglobin by modification of manual hemoglobin cyanide method.Red blood cells and other blood cells were counted electronically in systems based on light scattering technology.

A three part differential count assigns cells to categories usually designated (1) granulocytes or large cells (2) lymphocytes or small cells (3) mononuclear cells or middle cells

Platelets can be counted using same techniques of electrical or electro optical deflection as are employed for counting red cells. Platelets can be counted between two fixed thresholds for example between 2 and 20 fl.

Peripheral smear:

Ideal smear should be tongue shaped with 4cm long and 2 cm wide was made and stained by Giemsa staining which contain basically mixture of acidic eosin y and contain basic stain of azure B thionin

ESR is measured by Wintrobes method. The anticoagulation used is 3.8% trisodium citrate.

CRP is calculated using a latex CRP reagent which consists of aqueous suspension of polysterene particles sensitized with a globulin fraction from anti CRP serum.

Rheumatoid factor:

Rheumatoid factor of IgM class can be detected and measured quantitatively by testing the ability of patient serum to agglutinate carrier particles coated with IgG. Polysterene particles coated with human IgG are used in the latex slide test.

Bleeding time:

It is calculated using filter paper method.

Clotting time:

It is calculated using tube method

Serum ferritin

Microplates coated with anti human ferritin antibody will bind to ferritin from serum on the coated plates and non bound component is removed with washing. Then "antihuman ferritin horseradish peroxidase conjugate" is pipeted which binds to ferritin and excess is washed off. With chromogenic substrate of tetramethyl benzidine color develops and which is titrated by HCL. Amount of color developed is proportional to the concentration of ferritin which is measured by the optical density with a 450nm filter with micro plate reader.

DAS 28 score

Disease activity score is a composite score using tender and swollen joints count , ESR and patients global assessment activity using a 100 mm visual analogue scale.

$$\mathbf{DAS28=0.56\ \sqrt{(no.of\ tender\ joints)} + 0.28\ \sqrt{(no.of\ swollen\ joints)} +0.70\ log(ESR)+0.014(global\ assessment\ in\ mm).}$$

Classification

Mild ≤3.1

Moderate 3.2-5.1

Severe >5.1 (Minimum score :0; Maximum score : 9)

Parameters used in Disease activity score:

(1) Total 28 joint count for tenderness

(2) Total 28 joint for swelling

(3) ESR in mm in first hour

(4) Patient assessment of global health using a 100mm visual analogue scale ranging from 0(very good) to 100 (very poor)

28 joint counts:

(1) Shoulders (2)

(2) Elbows (2)

(3)Wrists (2)

(4)MCP for 4 fingers (8)

(5)MCP thumb (2)

(6) PIP for 4 fingers and thumb (10)

(7) Knees (2)

Data analysis was done using epidemiological information statistical software. Using the software the frequencies, mean , standard deviation and p values calculated with yate's test for qualitative variables

and kruskol walls chi square test for quantitative variables. p value <0.05 is taken as significant.

RESULTS AND ANALYSIS OF OBSERVED DATA

Table 1

Sex Distribution

Sl. no	Sex	N0. of patients	Percentage
1	male	9	20%
2	female	35	80%

In this study out of 44 cases , 35 are females and 9 are males.

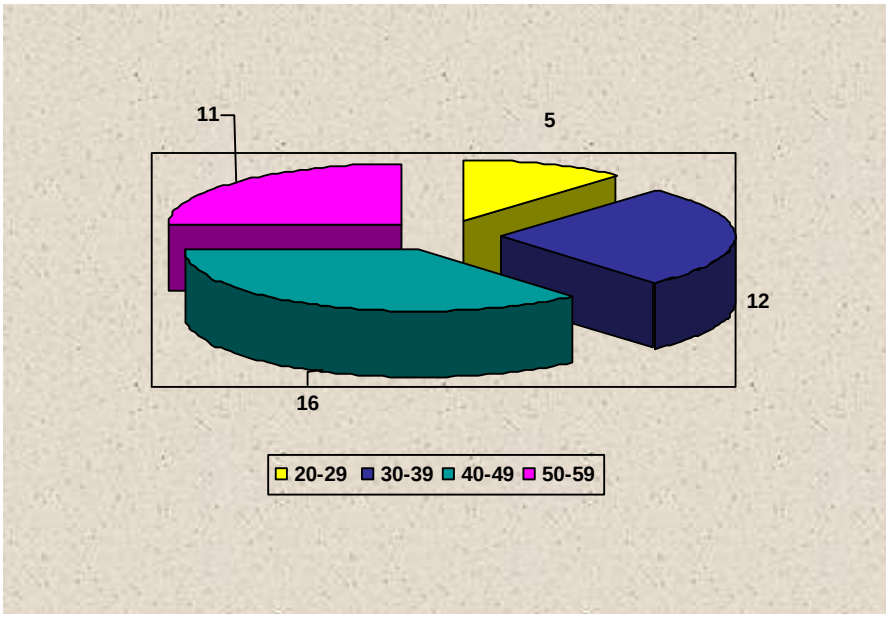
The female are 80% and males 20%.

Table 2: Age distribution

Age group	Cases	
	No.	%
20-29 years	5	11.4
30-39 years	12	27.3
40-49 years	16	36.4
50-59 years	11	25
Total	44	100
Mean	40.98 years	
S.D.	9.73 years	

The age of the patients from 20-60 years with an average of 40 years and more cases from 40- 49 years of age group.

AGE DISTRIBUTION



SEX DISTRIBUTION

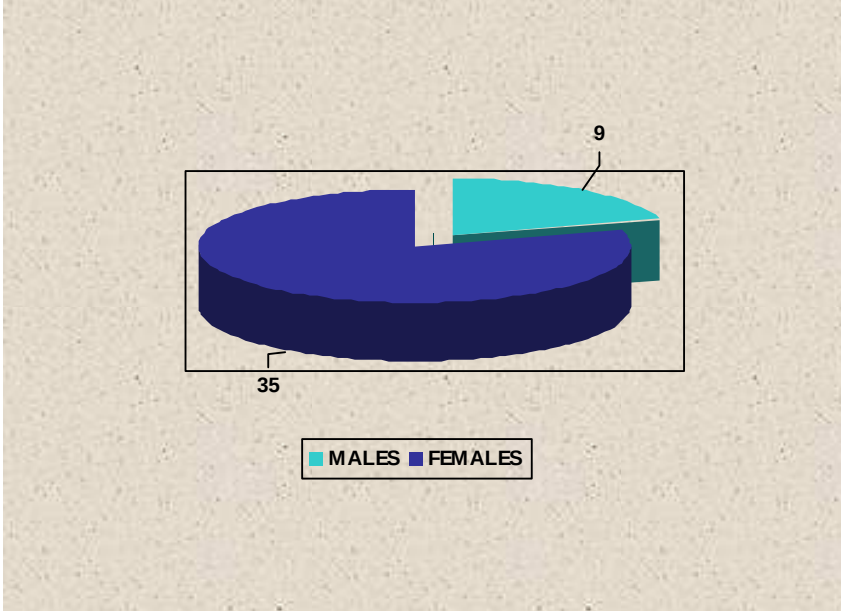


Table 3: DAS Score 28

DAS Score 28	Cases	
Mild (≤ 3.1)	2	4.5
Moderate (3.2-5.1)	23	52.3
Severe (> 5.1)	20	45.5
Total	44	100
Score Range Mean S.D.	2.75 -5.81 4.8 0.78	

This table shows that 2 people out of 44 (4.5%) had mild disease and 23 people (52.3%) has moderate disease. 20 people (45.5%) had severe disease. DAS 28 score ranges from 2.75 to 5.81 with a mean value of 4.8 with standard deviation 0.78

DAS 28 SCORE

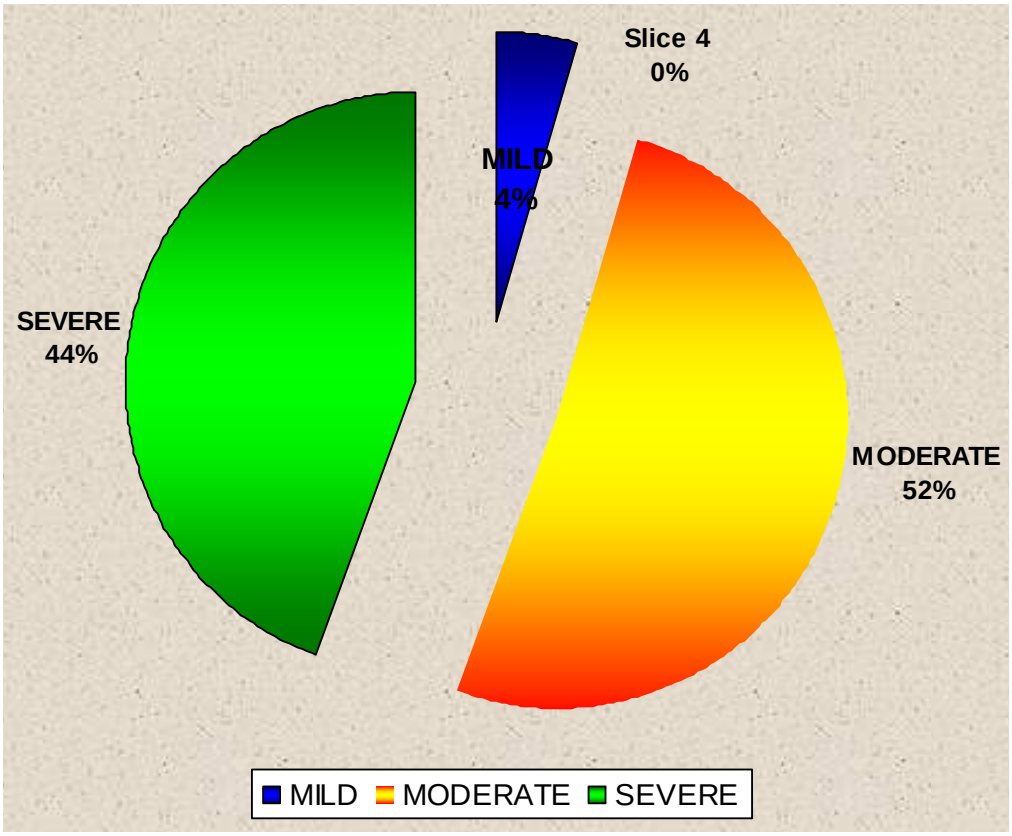


Table 4 : DAS 28 Score and Age

Age group	DAS 28 Score	
	Mean	**S.D.**
20-29	4.8	0.5
30-39	4.48	0.67
40-49	4.52	0.69
50-59	4.51	0.92
'p'	0.8108 Not Significant	

Analyzing the above data age group doesn't correlate with disease activity and DAS 28 score

Table 5
Duration of illness

	Duration of illness(in years)
Range	1 – 5
Mean	2.93
S.D.	1.13

The duration of illness ranged from 1 year to 5 years with an average of 2.98 years with a standard deviation of 1.13 years.

All 44 cases(100%)fulfilled the revised criteria of American rheumatism association for rheumatoid arthritis

All the cases distal joints involvement(distal interphalangeal joints are spared) 14 cases had both proximal and distal joint involvement(31%). Joint deformity were present in 22 cases (50%) rheumatoid nodules were present in 3cases(6%). Episcleritis was present in 3 cases(6%). Aortic Valve involvement was in 2 cases (4%)

Table : 6

Rheumatoid factor positivity

Rheumatoid factor	Cases	
	No.	%
%Positive	35	79.5
Negative	9	20.5

Rheumatoid factor is positive in 35 cases (79.5%) and negative in 9 cases (20.5%).

Serum proteins were normal . There was no reversal of albumin / globulin ratio and there was no hyperglobulinemia noticed in the study .Serum calcium and uric acid were normal in all patients.

27 patients showed radiological evidence of rheumatoid arthritis. No patients had splenomegaly or significant generalized lymphadenopathy.

Concomitant usage of NSAIDS 80% and corticosteroids 40% and methotrexate 4% was present.

Anemia is defined as <11gm in females and <12gm in males as in most of the studies

Table No 7

Anemia and rheumatoid arthritis

Sl. no		No of cases	percentage
1	Anemic	33	75%
2	Not anemic	11	25%

Among the 44 cases of rheumatoid arthritis 33 cases are anemic (75%) and not anemic in 11 case (25%)Mean hemoglobin level in patients was 10.67 ± 1.83

Table No 8

Anemia and rheumatoid factor positivity:

Anemic	26/33
Not anemic	6/11
P value	0.0305 significant

In patients who are anemic , number of rheumatoid factor positivity was 87% and in not anemic patients rheumatoid factor positivity was only 54%.

Mean Hb level in rheumatoid factor positivity was 9.11 gms$\pm$2.05s.d

Mean Hb level in rheumatoid factor negativity was 10.23$\pm$1.19s.d

When analysing the above values, anemia is one of the indicator of disease activity and severity of rheumatoid arthritis

Anemia and ESR and Rheumatoid factor positivity

Anemic and non anemic patients were comparatively studied with their erythrocyte sedimentation rate levels and seropositivity for rheumatoid factor.

Out of 33 patients 32 patients have elevated ESR out of which rheumatoid factor positive in 29 patients (90.6%) whereas in 11 non anemic patients 10 had elevated ESR of which only 6 are rheumatoid

factor positive (60%). The values suggest that the anemic patients have more elevation of ESR and percentage of rheumatoid factor positivity is also more in this group.

Anemia with disease duration

In 33 anemic patients 15 patients had more than 3yr duration (45%) whereas in non anemic 11 patients only 1 had (9%) had disease more than 3 year. When analyzing the above, the incidence of anemia correlated with the duration of disease.

Table 9 : DAS 28 Score and duration of disease

DAS 28 Score	**Duration of disease (in years)**	
	Mean	**S.D.**
Mild	2.0	-
Moderate	2.83	1.17
Severe	3.5	1.16
'p'	**0.0471 Significant**	

When analyzing the above charts DAS 28 score was correlated very well with duration of disease.

ANEMIA AND DURATION OF DISEASE

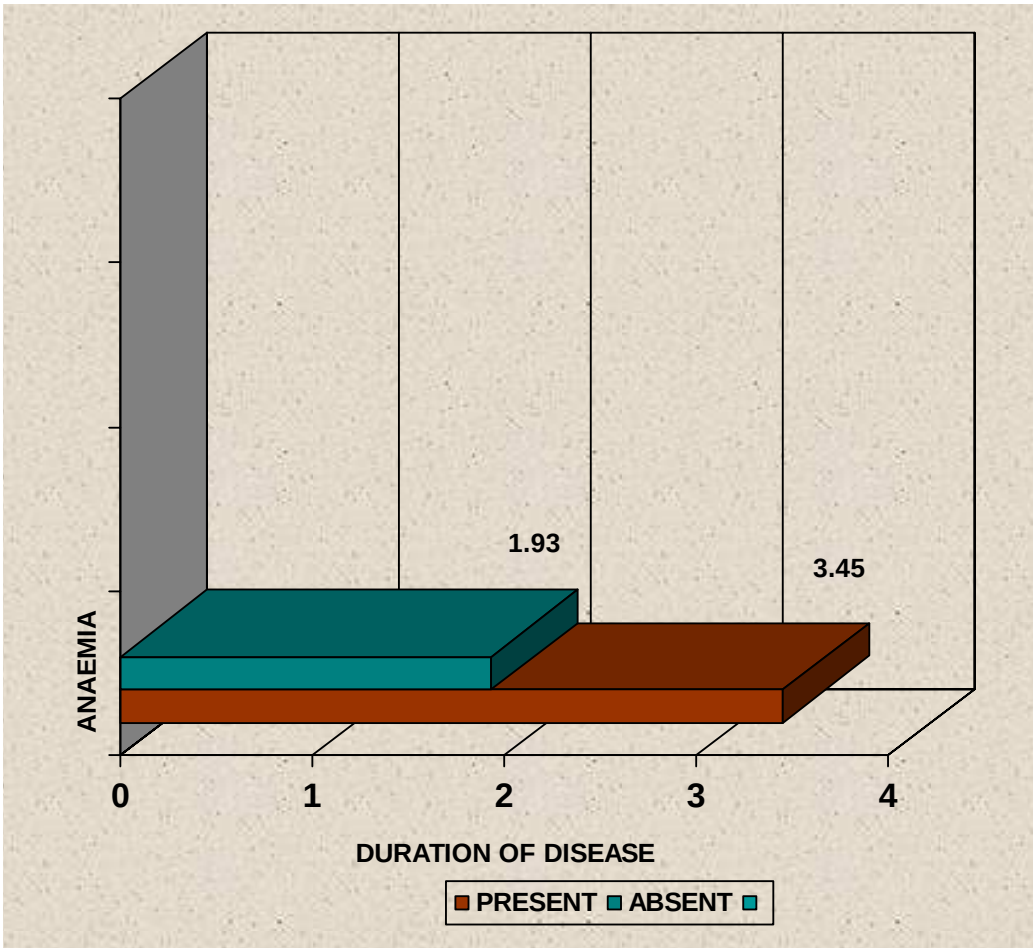


Table10: Anemia and DAS 28 Score

Anemia	DAS 28 Score	
	Mean	**S.D.**
Absent	4.32	0.91
Present	5.04	0.58
'p'	**0.0060** **Significant**	

When analyzing the data incidence of anemia correlated with activity of disease and anemic patients had higher DAS 28 score than non anemic patients. P value is significant.

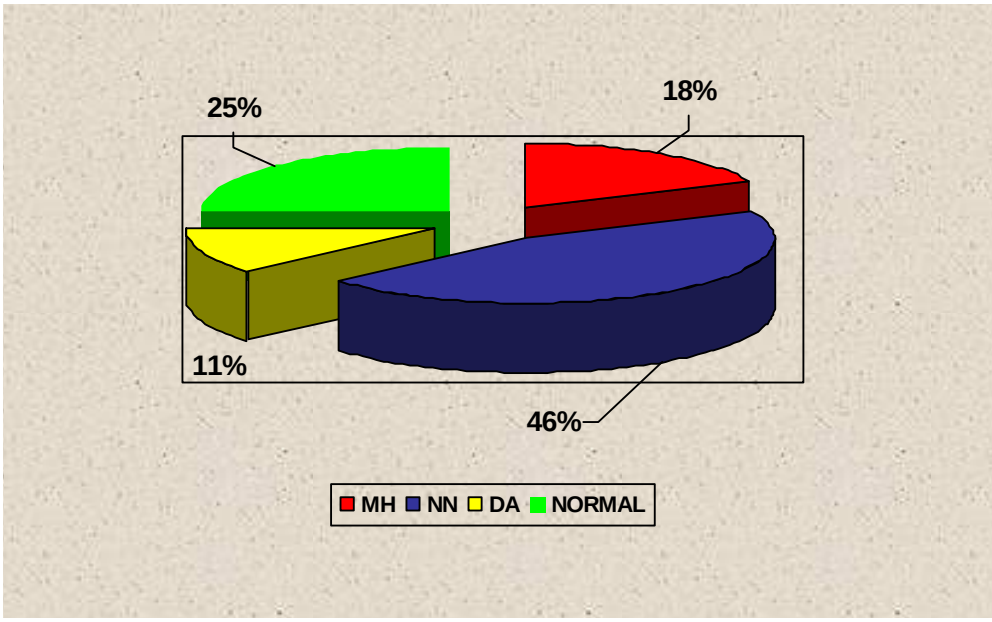
Table 11:Peripheral smear study

Types of anemia and rheumatoid factor positivity

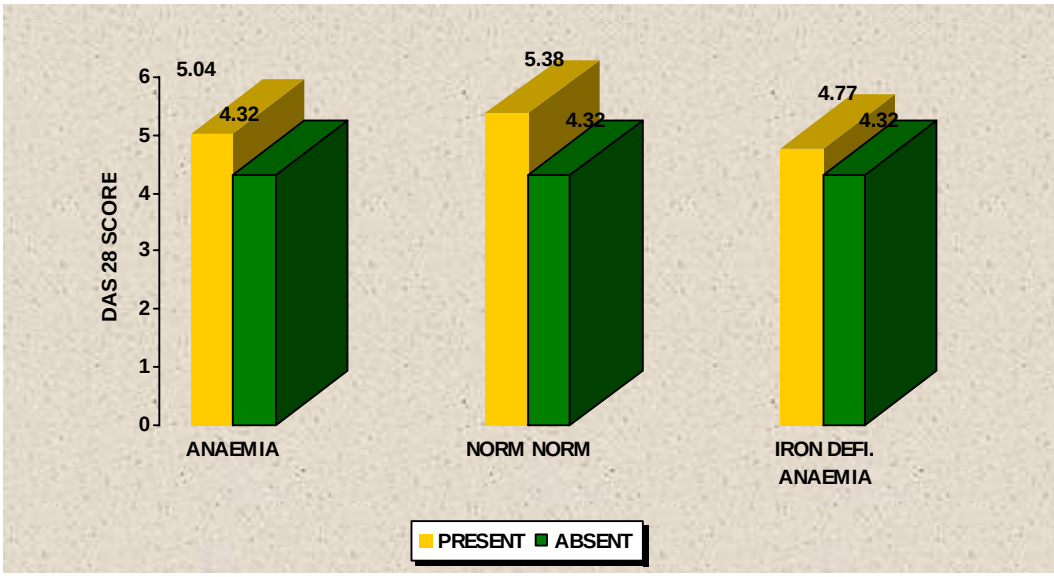
	No.of pts	**percentage**	**Rheumatoid positivity**	**percentage**
Microcytic hypochromic	8	18.2	8	100
Normocytic normochromic	20	45.5	17	85
Dimorphic	5	11.4	4	80
normal	11	25	6	54

When analyzing the above data 8 patients (18.2%)patients show microcytic hypochromic anemia . Out of 8 patients all shows rheumatoid factor positivity. 20 patients (45.5%) shows normocytic normochromic

PERIPHERAL SMEAR STUDY



ANEMIA AND DAS 28 SCORE



anemia. Out of 20 patients 17 patients (85%) were rheumatoid factor positive. 5 patients shows dimorphic anemia. Out of 5 patients 4 patients shows (80%) rheumatoid factor positive. But p value is 0.09 not significant. That means the type of anemia doesn't correlate with rheumatoid factor positivity .

Percentage of types of anemia in anemic patients:

Iron deficiency anemia 25%

Anemia of chronic disease 60%

Dimorphic anemia 15%

Table 12 : DAS 28 Score and Peripheral Smear Study

Peripheral Smear Study	**DAS 28 Score**	
	Mean	**S.D.**
Microcytic hypochromic anaemia	4.77	0.35
Normocytic normochromic anaemia	5.38	0.46
Dimorphic anemia	4.71	0.48
Normal	3.8	0.51
'p'	**0.0001 Significant**	

When analyzing the above data anemic patients has more DAS 28 score than not anemic patients. Patients with normocytic anemia that means anemia of chronic disease has high DAS 28 score (5.38) than iron deficient patients (4.77) p value is significant 0.001

DAS 28 SCORE AND TYPES OF ANEMIA

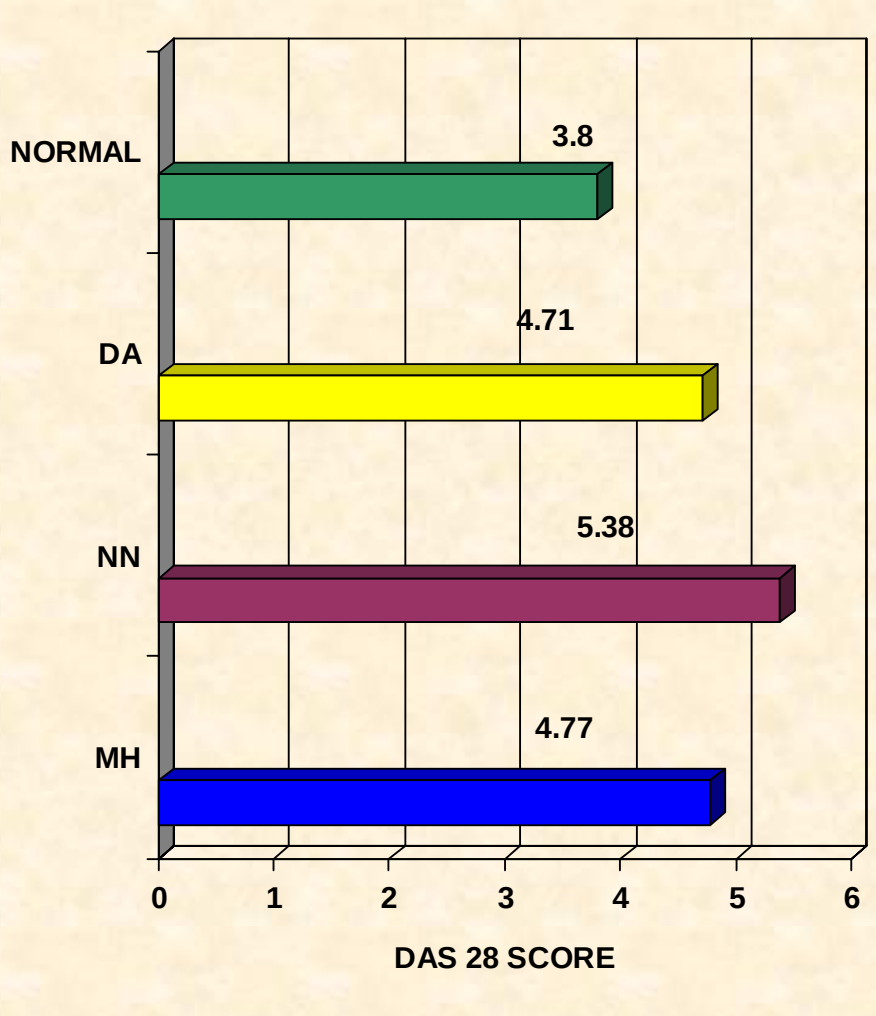


Table 13: DAS 28 Score and Rheumatoid Factor

Rheumatoid factor	DAS 28 Score	
	Mean	**S.D.**
Positive	4.98	0.61
Negative	4.01	0.53
'p'	**0.0025** **Significant**	

When analyzing the above data rheumatoid factor positive patients have higher DAS 28 score than rheumatoid factor negative patients

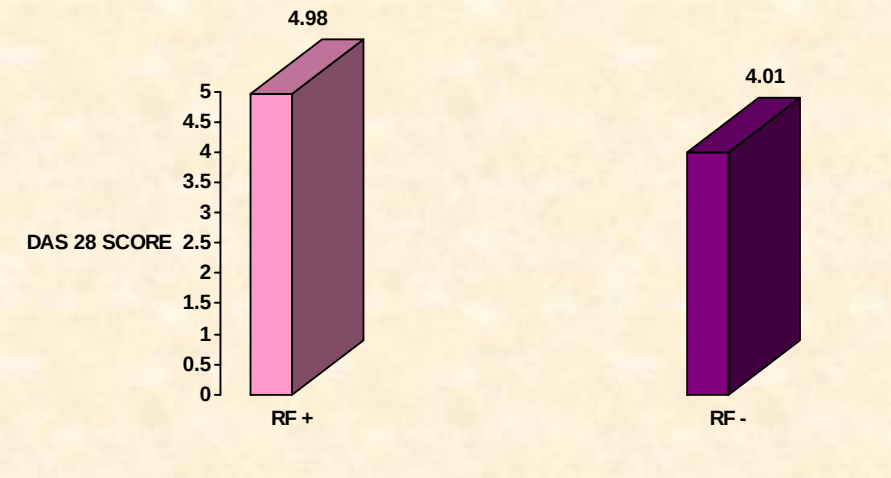
Table 14:

Serum ferritin and DAS 28 score

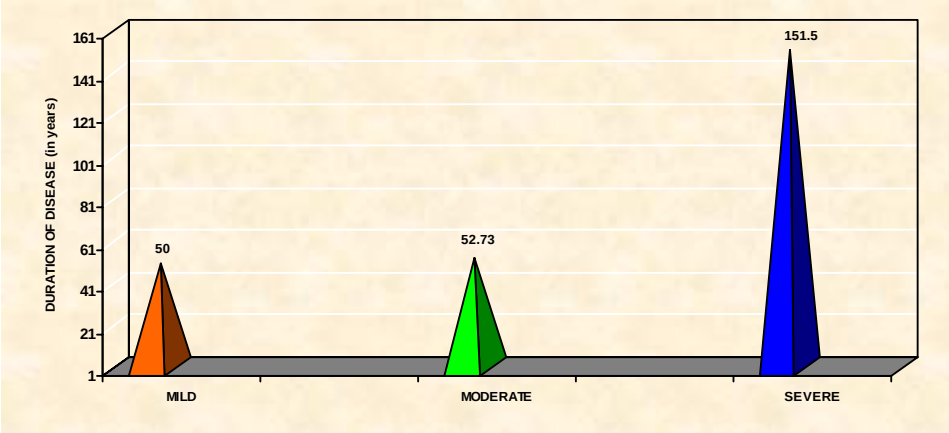
DAS 28 sore	Serum ferritin(mean &S.D)
Mild	50±24.5
Moderate	52.73±34.77
Severe	151.5±88.9
P VALUE	**0.001 significant**

The table shows that ferritin value correlates with severity of rheumatoid arthritis.

DAS 28 SCORE AND RHEUMATOID FACTOR



DAS 28 SCORE AND SERUM FERRITIN



Packed cell volume;

Mean value 34 ± 5.3

<20% -1 patient -2%

20-30% --9 patients -20%

30-40% -22 patients -50%

>40% -5 patients -11%

Table 15:

Clinical and laboratory features of anemic and non anemic patients:

	Anemic patients		**Not anemic patients**		
	Mean	**S.D**	**Mean**	**S.D**	**P value**
Tender joint count,	10.4	5.45	5.27	3.85	**0.004 significant**
Swollen joint count	6.5	3.78	3.28	4.82	**0.oo6 significant**
Visual analogue scale	66.10	53.1	31.2	43.3	**0.005 significant**
Hemoglobin	9.72	1.43	12.49	0.75	**0.0001 significant**
Mean corpuscular volume	78.27	8.87	86.42	5.3	**0.0036 significant**
Mean corpuscular hemoglobin	24.51	3.63	27.92	2.2	**0.0033 significant**
Mean corpuscular hemoglobin concentration	31.08	1.73	32.27	0.81	**0.0071 significant**

In this study MCV and Hb and MCH an MCHC has higher values in anemic patients than non anemic patients .p value was significant . In DAS 28 score we are using the variables like swollen joints and tender

joints and visual analogue scale and ESR. ESR already shows the highly significant correlation with anemia and rheumatoid factor positivity and disease activity. Swollen joints and tender joints and visual analogue scale with anemia correlation was highly significant. P value was <0.05

Table 16:

Clinical and laboratory features of iron deficient anemic and anemia of chronic disease patients

	IDA Patients		**ACD Patients**		**P value**
	Mean	**S.D**	**Mean**	**S.D**	
Tender joint count	5.54	3.57	10.55	7.40	**0.006 significant**
Swollen joint count	4.01	2.78	7.65	5.43	**0.006 significant**
Visual analogue scale	41.12	48.08	78.12	45.87	**0.002 significant**
hemoglobin	8.6	1.81	10.91	0.82	**0.0019 significant**
Mean corpuscular volume	76.84	5.87	83.97	5.0	**0.002 significant**
Mean corpuscular hemoglobin	21.22	3.01	26.93	1.86	**0.003 significant**
Mean corpuscular hemoglobin concentration	29.8	2.01	31.83	1.3.	**0.0032significant**

This table tells that Hb , MCV and MCH and MCHC are lower in Iron deficiency anemia than anemia of chronic disease. Tender joint count and swollen joint count and visual analogue scale also lower in

iron deficiency anemia than anemia of chronic disease. P value was significant in all variables.

WBC count:

Out of 44 patients 13 patients have leucocytosis (29.5%). No patient had leucopenia.

Neutrophils:

50-70%- 8 patients -18%

>70%-36 patients -82%

Neutrophilia is present in 36 patients.

Lymphocytes:.

<20%-10 patients -23%

20-30%-18 patients -41%

>30%-16 patients -36%

Lymphopenia is present in23% of patients.

Eosinophils:

12patients (27%) have eosinophils >6%. All 12 patients are rheumatoid factor positive.

No patients had immature cells or large granular lymphocytes.

Thrombocytes

Table 17:
Platelet count and rheumatoid factor positivity

	No. of patients	Rheumatoid factor positive	percentage
>4 lakhs	14	12	85%
<4 lakhs	30	23	76%

When analyzing the above data thrombocytosis indicates disease activity. P value was significant 0.04.

Table 18

Relationship between DAS 28 Score and other parameters

Parameter	Value for cases with DAS 28 Score						'p'
	Mild		Moderate		Severe		
	mean	S.D.	mean	S.D.	mean	S.D.	
PLT	2.75	1.35	3.49	0.93	4.09	0.97	**0.0046 Significant**
Eosinophil.	1	1.4	2.39	3.25	6.7	5.76	**0.0001 Significant**
CRP	6.8	5.5	23.13	24.5	53.85	19.9	**0.0006 Significant**
Ferritin	50	2.3	52.73	34.7	151.5	88.9	**0.001 significant**
ESR	25	14.1	43.24	12.2	67.25	25.5	**0.0001 Significant**

When analyzing the above data ESR and CRP, platelet count and eosinophil count and ferritin are well correlated with disease activity . that means if ESR and CRP and platelet count and eosinophil and ferritin was high ,disease activity and DAS28 score was also high. All the other parameters doesn't correlate with DAS 28 score.

Bleeding time ;

Normal in all patients.

Clotting time

Normal in all patients.

No patients had features of hyperviscosity syndrome and no patient had a features of Felty syndrome and no patient had a feature of pure red cell aplasia and no lymphoma and leukemia.

DISCUSSION

Rheumatoid arthritis is a chronic, systemic, inflammatory disorder of unknown etiology that is a pattern of diarthrodial joint involvement. Its primary site of pathology is the synovium of joints. The rheumatoid factor positivity and extra articular manifestations commonly accompany the joint disease , but arthritis is the major manifestation.

In our study we selected 44 cases of rheumatoid arthritis on random basis as per the American rheumatism association guidelines 1987.

The sex distribution in this study , is predominantly affects females in a ratio of 4:1.According to Harrison 17th edition, API text book of medicine that the women are affected 3 times more than male. In this study males are 20% and females are 80%. Doran Mf, Ponal Gr *et al* [35] in his study males are 26.9% and females are 73.1%. Mean age is 58.5 years. In our study is 40.98 years.

Alamonsky, Yougari , Drosos et al [36] in his study the risk of developing disease is greatest between 40 and 50 years. In our study the risk also is between 40 to 49 years around 36.4 %.

Navarocaro Gregio *et al* [37]and Abach, R.R. Buchanan et al [38] in their study the severity of disease is in positive correlation with duration

of disease. In this study patients who had disease more than 3 year have more DAS 28 score .The p value is significant and this shows that duration of disease is directly proportional to the severity of disease.

B.Fleeb, L.Andel, J.Sautner et al [39], in their study the mean DAS 28 score was 4.23±1.2 .In this study mean DAS 28 score was 4.8±0.78. Toshihisomatsui et al [40] in their study reported 11.6% patients had mild DAS 28 score and 51.4% has moderate DAS 28 score and 37.4% patients had severe DAS 28 score. In this study patients with mild DAS 28 was around 4.5% and moderate score was 52.3% and severe 45.5% . This states that most of the patients are in moderate severity.

Card Richard *et al* [41] in his study rheumatoid factor positivity was 80% and negativity 20%. In this study RF positivity was 79.5% and while 20.5% RF was negative . The ratio of rheumatoid factor positivity to negativity is 4:1

Tracey Houston *et al* [42] in their study ,mean hemoglobin in rheumatoid arthritis patients was 9.57 gm% and in this study mean Hb is 10.6gm%.

M. Kar, S. Roy *et al* [43] in their study, mean hemoglobin level in rheumatoid positivity patients was 9.57gm% and 10.45gm%. among rheumatoid negative patients. In this study mean Hb level in rheumatoid factor positive patients is 9.11±2.05 and mean Hb among rheumatoid factor negative is 10.23±1.19 gm%.

D.J. Borah ,Farhis Iqbal *et al* [44] in their study ,out of 20 anemic patients 18 patients were rheumatoid factor positive (90%) and in non anemic patients out of 11 patients 6 patients (54%) were rheumatoid factor positive. In this study , out of 33 anemic patients 29 patients are rheumatoid factor positive (87%) and in non anemic 11 patients patients, 6 (54%) are rheumatoid factor positive. This states that anemia is very well correlated with rheumatoid factor positivity and disease activity.

Agarwal Sumeet *et al*[45] in their study, mean DAS 28 score in non anemic patients was 3.83 compared to anemic patients which was 5.13. In this study in non anemic patients mean DAS 28 score is 4.32 and in anemic patients 5.04. D J Borah , Farhis Iqbal *et al* [44] in their study, in non anemic patients mean DAS 28 score was 4.76 while anemic patients it was 6.85.This states that anemic patients have more DAS score and disease activity than non anemic patients.

Distinguishing ACD from iron-deficiency state in the setting of chronic inflammatory process is difficult. Serum ferritin is a reliable parameter to predict bone-marrow iron stores in uncomplicated anemic states.

However, in chronic inflammation, ferritin is increased as a part of acute-phase reaction and hence determining a cutoff level to differentiate between iron-replete and depleted state becomes difficult. The value of serum ferritin indicative of iron-deficient state in various studies range

from 30 to 70 μg/l [46, 47]. We have taken a value of 50 μg/l as an indicator of iron-deficiency in our study as it has been shown to have a specificity of 81% and sensitivity of 100% [23]. Bone-marrow iron staining is considered as the gold-standard in such conditions. However, the procedure is invasive, time-consuming and expensive. Two recent studies have shown that absent bone-marrow stainable iron may not truly be representative of iron deficiency (48).

Soluble transferrin receptors (sTfR) level has been found to be useful in distinguishing iron deficiency from ACD as it does not behave as an acute-phase reactant. A high sTfR level although indicates iron-deficient erythropoiesis, it does not necessarily indicate iron deficient state. A serum ferritin of 50 μg/l is as sensitive as sTfR in predicting iron-deficiency [23]. However a combination of high sTfR (> 2.5 mg/l) and low ferritin (< 50 μg/l) levels or a ratio of sTfR to log ferritin gave higher specificity (> 90%) than either of them alone in predicting IDA in RA [23]. It may also help differentiate patients with mixed anemia from pure ACD which is probably the most common in patients with RA.

Agarwal Sumeet et al (45), in their study, of rheumatoid arthritis patients with iron deficiency anemia DAS 28 score was 4.7 and in patients with anemia of chronic disease DAS 28 score was 5.69 . In this study patients with iron deficiency DAS 28 score was 4.77 and in

anemia of chronic disease was 5.38. The p value is significant(0.001) .This states that DAS 28 score is higher in ACD than in IDA.

	Anemic patients %
This study	75%
Peters et al [49]	64%
Baer etal[12]	76%
Remacha et al [51]	55%
Hotchberg et al [52]	40%
Baer et al [53]	27%
Segal et al [54]	49%

Anemia was defined as hemoglobin level of 11 g/dl in female and 12 g/dl in male patients the prevalence reported in RA patients from Western countries [49] which varies from 33.3 to 59.1% although the cutoff hemoglobin value used to define anemia in these studies was higher than our study.

Most of the above studies used a definition of anemia close to that suggested by World Health Organization (WHO) while we have used a definition lower than that. Despite this we find that the prevalence of anemia is considerably higher. If the WHO criterion had been used (men Hb 13 g/dl and women 12 g/dl) then the prevalence of anemia in our cohort would be 84.1%.

This is possibly related to high background prevalence of anemia in general adult Indian population as well as poor access to medical care leading to poor disease control of RA.

Types of anemia and rheumatoid arthritis

	Peter et[49]	JK sci [44]	Agarwal[45]	This study
ACD	65%	60%	51.6%	60.5%
IDA	23%	40%	48.4%	24.5%
Dimorphic	12%	---	----	15%

In this study iron deficiency anemia patients are less (24.5%) because iron deficient anemia with inflammation(Dimorphic anemia)is included separately(15%) and there is a probable folic acid and/or Vit B12 deficiency .

Microcytosis (<80 femtolitre) in this study is 27% patients among rheumatoid arthritis patients .Alexander *et al* [8] in their study showed 30% prevalence of microcytosis .

Hypochromia (less than 26pg) is present in 38% of rheumatoid arthritis patients in this study. Caris J Bastley et al [55] reported 50% hypochromia in their study.

Kadir *et al* [56] in his study, mean ESR was 36.6$\pm$23.5 .In their study ferritin was 121.3 $\pm$34.2 ,and ESR ,CRP and ferritin showed significant correlation with disease activity as well as DAS 28 score . The p value was significant <0.001. In this study mean ESR is 53.5$\pm$22.3 and ferritin is 99.5$\pm$80.93 and ESR, CRP and ferritin are very

well correlated with disease activity and the p value was 0.006 for ESR and 0.0001 for CRP and ferritin respectively .

Sumeet Agarwal *et al* [45] and D J Borah , Fahler Iqbal *et al* [44] in their study, the variables used in calculating DAS 28 score like tender joint count , swollen joint count , ESR and visual analogue scale was correlated significantly. The p value was more significant in anemic patients than non anemic patients .In this study also all the 4 variables shows high significance in anemic patients compared to non anemic patients.

Agarwal *et al* [45] in their study; tender joint count , swollen joint count , ESR and visual analogue scale in patients with anemia of chronic disease showed higher value than iron deficiency anemic patients. Similar results were obtained in all the variables showing higher significance in patients with anemia of chronic disease than iron deficiency anemic patients in this study. The p value is significant.

Hutchuson *et al* [57] and Dulaguist *et al* [58] in their study thrombocytosis in rheumatoid arthritis was 52% and 60% respectively Alof Selross et al [59] in their study ;thrombocytosis was present in 33% of patients and it correlated with disease activity. In our study thrombocytosis is present in 31%. Patients with thombocytosis have higher DAS 28 score with significant p value .

M.Kar , S.Roy *et al* [43] in their study ;eosinophilia was present in 20.45% of rheumatoid arthritis patients. R J Windchester *et al* [60] and Short Bauer *et al* [61] in their study eosinophilia was present in 40% and 10% of patients of rheumatoid arthritis respectively which correlated with disease activity . In this study eosinophilia is present in 27% patients which correlated very well with DAS 28 score and disease activity with significant p value .

M.Kar S.Roy *et al*[43] in their study leucocytosis was present in 20.7% and leucopenia was present in 11.1% patients and lymphopenia was present in 6% patients. In this study 29.5% patients have leucocytosis. None of the patient had leucopenia .Lymphopenia is present in 23% patients. Leucopenia is lacking in this study because we didn't take patients with longer duration disease for study group and lymphopenia is present higher than M.Kar S.Roy study ,probably due to steroid therapy .

Luis A, Toro Jimenez *et a*l [62] in their study of 214 rheumatoid arthritis patients 12 patients had M spike with mean age of M spike being 69 years. IgG gammopathy was present in 50% patients. 1 patient was diagnosed as multiple myeloma and 1 patient had a primary lymphoproliferative disorder . One patient diagnosed as a T cell

leukemia and 4 patients diagnosed as a myelodysplastic syndrome and 4 patients were diagnosed to have monoclonal gammopathy .

In this study no patient has hyperglobulinemia and myelodysplastic syndrome, leukemia or lymphoma or multiple myeloma and no patient had a large granular lymphocytosis syndrome.

Agarwal Sachdev *et al* [63] reported pure red cell aplasia and immune thrombocytopenia. In this study no patients had thrombocytopenia and pure red cell aplasia. No patient had decreased red cell distribution width .

Abach , R,R,Buchnan *et al*[38] reported a hyperviscosity syndrome in rheumatoid arthritis. In this study no patient had symptoms suggestive of hyperviscosity syndrome

Paraiaz , Fayaz *etal* [33] study none of the patient of rheumatoid arthritis had evidence of bleeding and DIC. Only isolated abnormalities of coagulation was present. Protein C and protein S was low in 1case each while decreased factor VIII level was detected in 5 cases. Hypofibrinogenemia was demonstrated in 1 case. All patients had normal factor IX level. In this study no patient had evidence of bleeding and abnormal bleeding time as well as clotting time.

CONCLUSION

(1) Sex ratio of females to males is this study is 4:1

(2)The risk of developing disease is greatest between 40 to 49 years .

(3) Rheumatoid factor positivity is 80% and rheumatoid factor negativity is 20%

(4)The prevalence of anemia in rheumatoid arthritis patients is 75%

(5) In rheumatoid factor positive patients mean Hb values is less (9.11gm%) compared to rheumatoid factor negative patients (10.23gm%). Iron deficiency anemia patients mean Hb is lower(8.6gm%) than in anemia of chronic disease is (10.9gm%).

(6)The prevalence of rheumatoid arthritis according to DAS 28 score categories in decreasing order are moderate 52.3% , severe 45.5% and mild 4.5%.

(7) Anemia is very well correlated with rheumatoid factor positivity , disease activity (DAS 28 score) ,duration of disease and ESR.

(8) Microcytic hypochromic anemia (iron deficiency anemia) is present in 25% of anemic patients and anemia of chronic disease (normocytic normochromic) anemia is present in 60% of anemic patients and dimorphic anemia in 15% anemic patients.

(9)Patients with anemia of chronic disease have higher disease activity (DAS 28 score) than iron deficiency anemia patients in rheumatoid arthritis .

(10) Rheumatoid factor , ESR, CRP and ferritin positively correlates with DAS 28 score significantly..

(11) Thrombocytosis is present in 31% of patients and eosinophilia is present in 27% of patients and very well correlated with DAS 28 score.

(12) 29.5% have leucocytosis.while none of the patients have leucopenia . Lymphopenia is present in 20% of RA patients.

(13) Bleeding time and clotting time are normal among all patients .

SUMMARY

The study “Hematological Profile in Rheumatoid arthritis ” is a cross- sectional study conducted on patients visiting the outpatient Department of Rheumatology ,Government Rajaji Hospital , Madurai. Forty four patients with rheumatoid arthritis fulfilling the criteria of American rheumatologist association criteria (1987) were included in the study. Selected patients underwent clinical , and laboratory evaluation to detect the hematological abnormalities in rheumatoid arthritis patients and DAS 28 score was calculated which indicates the disease severity. It has been observed that anemia is the one of the indicator of disease severity and anemia incidence is more in South Indian patients than European Population. Anemia of chronic disease is more common compared to iron deficient anemia among rheumatoid arthritis patients . Anemia of chronic disease has higher DAS28 score than iron deficiency anemia as observed are in this study. It was also observed that thrombocytosis and eosinophilia also the indicators of disease severity which positively correlated with DAS 28 score; like ESR, CRP , rheumatoid factor and ferritin.

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GLOSSARY

ACD	-	Anemia of chronic disease
CRP	-	C reactive protein
DAS	-	Disease activity score
DIC	-	Disseminated intravascular coagulation
ESR	-	Erythrocyte sedimentation rate
Hb	-	Hemoglobin
IDA	-	Iron deficient anemia
MCV	-	Mean corpuscular volume
MCH	-	Mean corpuscular hemoglobin
MCHC	-	Mean corpuscular hemoglobin concentration
PCV	-	Packed cell volume
PDW	-	Platelet distribution width
RA	-	Rheumatoid arthritis
RBC	-	Red blood cell
RF	-	Rheumatoid factor
RDW	-	Red cell distribution width
sTfr	-	Soluble transferrin receptor
TIBC	-	Total iron binding capacity
WBC	-	White blood cell count
%sat	-	Percentage of saturation

PROFORMA

Name: age sex

Address: occupation

Symptoms duration

(1) joint pain
(2) joint swelling
(3) fever
(4) morning stiffness
(5) edema
(6) loss of weight
(7) loss of appetite
(8) muscle pain
(9) back pain
(10) recurrent infection
(11) easy fatiguability
(12) breathlessness
(13) bleeding manifestations

Past History

HT DM IHD TB

Personal history:

Smoking alcohol veg/nonveg

Education urban / rural

Drug H/O

Nsaids/ diuretics/steroids/immunosuppressants/ others

G/E pallor lymphadenopathy jaundice

Nodule ulcer dry eyes

Rash pedal edema skin infections

Temp PR RR BP

		Joint swelling				Joint tenderness			
		right		left		right		left	
		UL	LL	UL	LL	UL	LL	UL	LL
PIP	1								
	2								
	3								
	4								
	5								
MCP	1								
	2								
	3								
	4								
	5								
wrist									
shoulder									
elbow									
knee									

TM joint SI joint spine
cervical

Thoracic

Lumbosacral

Stage of progression Xray wise stage; Joint deformities:

Swelling joint count:

Tender joint count

Visual analogue scale 0----------100

Functional class I II III Iv

Other system examination

CVS	normal	pericarditis	myocarditis	endocarditis	Aortitis
RS	normal	Pleual effusion	ILD	bronchilitis	
abdomen	Normal	hepatomegaly	splenomegaly	ascites	
eyes	normal	sicca	scleritis	episcleritis	scleromalacia
CNS	normal	Cranial nerve palsy	Peripheral neuropathy	quadriplegia	

Psychiatry anxiety / depression

Investigations:

RBc WBC Platelet
HB DC MPV
HCT
MCV
MCH
MCHC
RDw

Serum ferritin
TIBC
Serum iron

Peripheral smear

BT CT

ESR	CRP	Rh factor
Sugar	urea	creatinine
Total protein	albumin	globulin

DAS 28 score

Master Chart

s no	age	sex	hb	rbc	pcv	mcv	mch	mchc	rdw	plt	wbc	neurto	eosin	lymphocy	mono	crp	mpv	ferr	iron	tibc	%sat	esr	das 28	rf	duratn(y)	ps
1	50	f	6.8	2.72	21.4	78.5	25	31.8	18.2	5.09	12,100	71	8	17	3	13.8	7.3	5.5	13	370	4	45	4.38	p	3	mh
2	29	f	9.4	4.41	30.1	68.2	21.4	31.4	18.8	4.38	7100	71	3	19	7	55.7	7.6	7	9	271	3	115	5.17	p	4	mh
3	35	f	12.4	4.32	37.7	87.4	28.8	33	16	3.84	20900	64	1	32	3	16.3	8.3	63.2	47	314	15	40	4	p	1	n
4	51	f	8.6	4.36	29.6	68	19.7	29	20.9	4.66	11500	76	10	9	5	90.5	9	31.8	5	323	2	60	5.12	p	2	mh
5	55	f	10.7	4.06	33.5	82.3	26.3	32	20.5	3.88	12200	72	1	22	5	9.4	6.9	48.5	36	362	10	30	4.24	n	3	da
6	47	f	10.4	3.52	31.9	90.8	29.7	32.8	15.3	5.14	6300	56	15	29	0	69.4	9	216	44	330	13	50	5.71	p	4	nn
7	20	f	10	3.78	30.3	80.4	26.5	32.9	15.1	5.16	9700	61	9	28	2	63.9	9.4	206	20	324	6	75	5.69	p	2	nn
8	58	m	11	3.72	35.4	95.1	30.2	31.7	14.8	5.97	9000	53	20	27	0	63.1	8.4	90	205	354	58	50	5.57	p	3	nn
9	52	m	11.6	4.18	35.2	84.2	27.7	32.9	16.4	3.09	11200	75	3	17	5	11	8.2	63	139	400	35	15	4.42	n	2	nn
10	59	m	12.1	3.74	35.8	95.8	32.3	33.7	13.9	2.75	5200	68	0	27	5	2.9	8.1	119	79	256	31	15	2.75	p	2	n
11	48	f	13.6	4.45	40.8	91.7	30.5	33.2	14.1	2.36	9400	69	3	24	4	2.9	9.4	78	64	358	18	35	3.75	n	2	n
12	56	m	11.4	4.5	41	91.2	29.8	32.7	14.6	4.02	15600	70	10	16	4	41.3	8	306	46	302	15	115	5.74	p	4	nn
13	34	f	10.7	4.45	37.1	83.3	26.6	32	16.6	3.84	10100	65	0	29	6	52.9	7.8	180	34	558	6	100	5.21	p	1	nn
14	47	f	10.8	4.58	38.4	83.8	26.7	31.9	17	3.47	9200	60	15	24	1	62.9	8.9	175	158	407	39	80	5.46	p	3	nn
15	54	f	10.7	4.01	33.5	83.6	26.6	31.9	15.6	4.62	10500	87	0	13	0	75.9	7.3	50.2	28	350	8	40	4.38	n	4	nn
16	50	f	10.7	4.18	34.9	83.5	25.6	30.6	17.2	4.12	9400	60	7	31	0	61.3	9.4	221	19	502	4	60	5.81	p	4	nn
17	40	m	11.3	4.18	35	83.7	27	32.2	15	4	5600	60	2	32	0	22.6	9.1	116	20	371	5	40	5.67	p	2	nn
18	36	f	8.3	4.08	29.2	71.5	20.4	28.6	20.4	5.45	14900	70	11	14	5	61.3	9.1	5	6	409	1	65	5.22	p	5	mh
19	41	m	12.8	5.11	41.6	81.4	25.1	30.8	17.5	4.03	10900	67	2	26	5	8.8	7.3	97	27	406	7	45	3.85	n	2	n
20	35	f	7	4.31	26.8	62.3	16.2	26	22.6	2.68	5200	91	1	7	1	50.1	9.6	5.8	6	472	1	40	4.69	p	5	mh
21	35	f	10	4.81	37.1	83.3	26.7	26.6	16.2	2.65	6800	58	3	36	3	31.3	9.4	209	46	425	11	40	5.41	p	4	nn
22	40	f	11.8	4.84	38.4	79.3	24.4	30.8	17.1	4.4	16700	64	8	24	4	60.3	8.4	5.2	150	452	33	90	5.24	p	3	da
23	48	m	14.2	4.77	43.9	92.1	29.7	32.3	15.2	3.3	10600	78	0	17	5	2.9	7.4	67	76	377	20	35	3.08	n	2	n
24	40	m	12.7	4.65	39.5	89	28.6	32.2	13.7	2.7	8600	77	1	17	4	2.9	8.8	88	26	349	7	35	3.3	n	1	n
25	40	f	9.2	5.18	30.9	59.6	17.8	29.8	23.9	3.67	7300	67	1	26	6	2.9	8.9	19.7	28	358	8	45	4.21	p	3	da
26	50	m	12.1	4.01	37.6	93.8	30.1	32	15.9	3.65	9200	68	1	28	3	3.1	9.5	73	41	365	11	45	4.03	p	2	n
27	40	f	13.3	4.57	39.5	86.2	27.1	31.4	17	4.56	9200	57	2	32	9	80.7	7.5	68.7	40	326	12	60	4.69	p	4	n
28	20	f	10.8	4.62	41.4	89.6	28.8	32.1	13.9	3.23	12900	79	1	17	3	65	8.7	163	55	275	20	80	5.71	p	1	nn
29	24	f	12.4	4.83	38.8	80.2	25.7	31.8	17.1	3.7	8600	66	1	29	4	2.9	8	7.8	31	477	6	60	4.37	p	3	mh
30	47	f	10.9	2.12	35.1	85.3	27.3	32	18.3	3.22	10100	59	1	35	5	77.1	7.9	277	19	286	7	45	5.63	p	4	nn
31	25	f	11.9	4.23	36	85	28.1	33.1	12.2	3.34	10000	58	1	40	1	3	7.3	93	78	380	20	60	5.33	p	2	nn

32	40	f	6	3	19.4	65	20	30.9	16.6	5.53	12400	50	15	32	3	60.7	7.9	47	39	280	14	65	5.1	p	3	da
33	40	f	12.3	4.54	38.3	84	27	32	13.5	2.56	10700	58	2	39	1	3.1	7.8	65	78	349	23	60	4.12	p	2	n
34	35	f	10.4	4.18	32.6	78	24.8	31.8	14.7	5.3	14900	62	10	27	1	63	7.2	233	20	370	6	65	5.43	p	4	nn
35	36	f	10.3	3.75	31.9	85	27.5	32.3	12	2.4	8000	58	1	41	0	41	7.1	53	47	420	11	35	4.62	n	3	nn
36	34	f	10.6	4.26	34	80	24.8	31.1	15.8	2.52	11200	51	6	36	1	51	8.3	175	44	330	13	70	5.53	p	4	nn
37	34	f	10.9	4.44	35.4	80	25.1	31.5	14.2	4.66	16300	72	2	26	0	10.7	6.6	110	79	256	31	35	4.78	n	2	nn
38	40	f	8.7	3.87	28.2	73	22.4	30.7	15.6	2.62	7600	62	2	35	1	45.8	7.1	11.4	139	300	46	60	4.74	p	5	da
39	50	f	10.2	3.89	30.6	80	26.3	33.4	12.6	3.14	5000	55	1	43	1	51	7	160	28	350	8	40	5.78	p	2	nn
40	46	f	9	4.49	30.9	69	20.1	29.1	19.2	3.33	10800	55	2	42	1	25	8	5	13	420	3	45	4.63	p	4	mh
41	40	f	9.9	4.34	31.7	73	22.7	31.1	15.7	3.72	9800	55	1	43	1	30.3	6.9	162	46	340	14	45	5.69	p	4	nn
42	30	f	12.6	4.98	28.3	80	26.2	32.8	12.5	2.5	15300	70	0	27	3	5	8	59	47	318	15	35	4.03	p	3	n
43	37	f	7.3	3.42	23.7	69	21.3	30.7	15.1	3.18	5600	68	5	26	1	37.1	8.1	7	10	340	3	55	4.57	p	4	mh
44	35	f	10.3	4.11	31.6	80	25.1	32.7	12.9	2.41	5600	58	1	40	1	20	7.5	120	64	356	18	35	3.77	p	2	n